

IN THE CLAIMS

Please amend the claims as follows:

Claim 1. (Currently Amended): A method for producing a heterologous RNA of interest, comprising:

(1) transforming mitochondria of a yeast cell lacking mitochondrial DNA with a mitochondrial transcription vector that comprises at least one copy of the DNA encoding said heterologous RNA of interest under the control of regulatory element(s) for mitochondrial transcription, and a mitochondrial transformation reporter gene or a fragment of said reporter gene,

(2) identifying a yeast mitochondrial transformant obtained in (1) which has mitochondria comprising the mitochondrial transcription vector but no mitochondrial DNA, wherein the only RNAs which are produced in said mitochondria by said mitochondrial transcription vector, are the heterologous RNA of interest alone, when the reporter gene or the fragment thereof are not transcribed, or the heterologous RNA of interest and the transcript of the reporter gene or reporter gene fragment, when said reporter gene or fragment thereof are transcribed that has incorporated the DNA of interest;

(3) culturing the yeast mitochondrial transformant selected in (2),

(4) isolating the mitochondria from the yeast mitochondrial transformant obtained in (3), and

(5) extracting and purifying the heterologous RNA of interest from said mitochondria.

Claim 2. (Previously Presented): The method of claim 1, wherein said yeast cell lacking mitochondrial DNA is a *rho*⁰ cell.

Claim 3. (Currently Amended): The method of claim 1, wherein said cell lacking mitochondrial DNA is obtained from a Δ *SUV3* or ~~*ADSS1*~~ strain.

Claim 4. (Cancelled)

Claim 5. (Previously Presented): The method of claim 1, wherein said DNA encoding the RNA of interest is under the control of a promoter and a transcription terminator that are functional in yeast mitochondria.

Claim 6. (Previously Presented): The method of claim 1, wherein said mitochondrial transformation reporter gene is a gene encoding one of the proteins of a yeast respiratory chain.

Claim 7. (Previously Presented): The method of claim 1, wherein said mitochondrial transcription vector comprises the sequence of an origin of replication of the mitochondrial DNA.

Claim 8. (Previously Presented): The method of claim 1, wherein the transformation according to (1) comprises the adsorption of said mitochondrial transcription vector onto metal microprojectiles and the projection of said microprojectiles onto said cells.

Claim 9. (Currently Amended): The method of claim 1, wherein (1) comprises the cotransformation of said yeast cells with said mitochondrial transcription vector and a vector that is replicative in yeast and that comprises a nuclear selection marker.

Claim 10. (Previously Presented): The method of claim 9, wherein said nuclear marker is an auxotrophic marker of said transformed cells.

Claim 11. (Previously Presented): The method of claim 1, wherein (2) comprises:
(a₀) crossing the yeast mitochondrial transformant obtained in (1) with a yeast tester strain of *rho*⁺ mit⁻ type,
(b₀) identifying a mitochondrial transformant which, once crossed, gives a diploid cell capable of growing on a non-fermentable medium, and
(c₀) repeating said crossing until isolated yeast colonies identified as being mitochondrial transformants carrying the mitochondrial transformation vector are obtained.

Claim 12. (Previously Presented): The method of claim 1,
wherein (1) comprises cotransformation of said yeast cell with said mitochondrial transcription vector and a vector that is replicative in yeast and that comprises a nuclear selection marker, and wherein (2) comprises:
(a₁) a first selection or preselection of the yeast cells by means of said nuclear marker,

by culturing in an appropriate medium, and

(b₁) a second selection from the yeast cell selected in (a₁), which comprises:

crossing the yeast mitochondrial transformant obtained in (1) with a yeast tester strain of *rho*⁺ mit⁻ type,

identifying a mitochondrial transformant which, once crossed, gives a diploid cell capable of growing on a non-fermentable medium, and

repeating said crossing until isolated yeast colonies identified as being mitochondrial transformants carrying the mitochondrial transformation vector are obtained.

Claim 13. (Previously Presented): The method of claim 1, wherein the isolation of the mitochondria, in accordance with (4) of the method, comprises lysis or grinding of said cell, and then at least two centrifugation steps, at speeds preferably of between 750 g and 12 500 g, and recovery of the final centrifugation pellet.

Claim 14. (Previously Presented): The method as claimed in claim 1, wherein (5) comprises:

eliminating the contaminating nucleic acids in the presence of appropriate buffers, the first buffer comprising at least one divalent ion-chelating agent, and the second buffer comprising an RNase and, optionally, a DNase,

lysing the mitochondria in the presence of at least one detergent and a divalent ion-chelating agent and within a pH range of between 7 and 8, and

isolating and purifying the RNA of interest.

Claims 15-20 (Cancelled)

Claim 21 (Currently Amended): A method for producing a heterologous RNA of interest comprising:

(1) transforming the mitochondria of a yeast cell lacking mitochondrial DNA with a mitochondrial transcription vector that comprises at least one copy of the DNA encoding said heterologous RNA of interest;

(2) identifying a yeast mitochondrial transformant obtained in (1) which has mitochondria comprising the mitochondrial transcription vector but no mitochondrial DNA, wherein the only RNAs which are produced in said mitochondria by said mitochondrial

transcription vector are the heterologous RNA of interest, and eventually the transcript of a mitochondrial transformation reporter gene or of a fragment of said reporter gene, only when said mitochondrial transcription vector comprises a mitochondrial transformation reporter gene or a fragment thereof that are transcribed that has incorporated the DNA of interest;

- (3) culturing the yeast mitochondrial transformant selected in (2);
- (4) isolating the mitochondria from the yeast mitochondrial transformant obtained in (3), and
- (5) extracting and purifying the heterologous RNA of interest from said mitochondria.

Claim 22. (Previously Presented): The method of claim 21, wherein the DNA encoding the heterologous RNA of interest is under the control of at least one regulatory element for mitochondrial transcription.

Claim 23. (Previously Presented): The method of claim 21, wherein said mitochondrial transcription vector comprises a mitochondrial transformation reporter gene.

Claim 24. (Previously Presented): A method for producing a heterologous RNA of interest, comprising:

- (1) transforming the mitochondria of a yeast cell lacking mitochondrial DNA with a mitochondrial transcription vector that comprises at least one copy of the DNA encoding said heterologous RNA of interest;
- (2) identifying a yeast mitochondrial transformant that has incorporated the DNA of interest;
- (3) culturing the yeast mitochondrial transformant selected in 2);
- (4) isolating the mitochondria from the yeast mitochondrial transformant obtained in 3), and
- (5) extracting and purifying the heterologous RNA of interest from said mitochondria, wherein said mitochondrial transcription vector comprises a fragment of a mitochondrial transformation reporter gene that is not transcribed.